A Section 508–conformant HTML version of this article is available at https://doi.org/10.1289/EHP7598.

Environmental Cadmium and Mortality from Influenza and Pneumonia in U.S. Adults

Sung Kyun Park,[1,2] Coralynn Sack,[3,4] Matti J. Sirén,[5] and Howard Hu[2,3,6]

[1]Department of Epidemiology, University of Michigan School of Public Health, Ann Arbor, Michigan, USA
[2]Department of Environmental Health Sciences, University of Michigan School of Public Health, Ann Arbor, Michigan, USA
[3]Department of Environmental and Occupational Health Sciences, University of Washington School of Public Health, Seattle, Washington, USA
[4]Department of Medicine, University of Washington School of Medicine, Seattle, Washington, USA
[5]Jorma Gallen-Kallela Memorial Research Library, Helsinki, Finland
[6]Department of Preventive Medicine, Keck School of Medicine, University of Southern California, Los Angeles, California, USA

Background: Environmental cadmium exposure is widespread. In humans, cadmium is poorly excreted, triggers pulmonary inflammation, reduces pulmonary function, and enhances lung injury by respiratory syncytial virus.

Objectives: We examined the association of cadmium burden with mortality related to influenza or pneumonia.

Methods: This prospective analysis of the National Health and Nutrition Examination Survey (NHANES) included 7,173 and 8,678 participants ≥45 years of age enrolled in NHANES-III and NHANES 1999–2006, respectively. Associations were evaluated between cadmium and mortality from influenza or pneumonia during a median follow-up of 17.3 y (NHANES-III, based on creatinine-corrected urine cadmium) and 11.4 y (NHANES 1999–2006, based on blood cadmium). Survey-weighted Cox proportional hazard models were used to compute hazard ratios (HRs) comparing the mortality of individuals at the 80th vs. the 20th percentile of cadmium concentrations.

Results: In NHANES-III, after adjustment for sex, race/ethnicity, education, body mass index, serum cholesterol, hypertension, and NHANES phase (or cycle), the HR comparing influenza or pneumonia mortality among participants with creatinine-corrected urinary cadmium in the 80th vs. 20th percentile was 1.15 (95% CI: 1.05, 1.26; $p = 0.002$) in the population as a whole and 1.27 (95% CI: 1.12, 1.43; $p = 0.002$) among never smokers. In NHANES 1999–2006, adjusted HRs for the 80th vs. 20th percentile of blood cadmium were 1.14 (95% CI: 0.96, 1.36; $p = 0.15$) for the overall population and 1.71 (95% CI: 0.95, 3.09; $p = 0.07$) in never smokers.

Discussion: Among middle-aged and older adults in the United States, higher cadmium burdens are associated with higher mortality from influenza or pneumonia. This raises the possibility that cadmium may worsen outcomes from COVID-19 infections. https://doi.org/10.1289/EHP7598

Introduction

There is an urgent need to identify modifiable risk factors that may predispose individuals with coronavirus disease 2019 (COVID-19) to developing worsening pneumonia, hypoxemia, or acute respiratory distress syndrome. This is particularly important given the continued absence of antiviral agents with proven efficacy and the uncertain timeline of vaccine development and availability. Mounting epidemiologic evidence has identified some personal characteristics that are associated with worse outcomes, including characteristics such as older age or preexisting chronic disease. Unfortunately, other than optimizing treatment for these chronic conditions, the associated opportunities for prevention are limited.

Environmental risk factors, however, are more amenable to prevention and can act through biological mechanisms relevant to COVID-19 as well as to other respiratory infection. One such factor that is has been observed to be a pulmonary immunotoxicant in several experimental studies and to which the general population has been widely exposed (from smoking and dietary sources) is cadmium (Cd) (Cohen 2004). Cadmium has been experimentally shown to be potent at inducing the *in vitro* expression and release of cytokines in human fibroblasts, epithelial cells, and macrophages following exposure to 7 μM Cd^{+2} for 7 h (Låg et al. 2010) and promoting lung inflammation and impairing macrophage-mediated immune function in studies of inhaled cadmium in rodents, leading to decreased resistance to pathogens (Koller 1998). Cadmium at noncytotoxic doses has been recently shown to disrupt tight junction integrity in a human air–liquid–interface airway tissue model, thereby impairing epithelial barrier function (Cao et al. 2015) and in a mouse model, cadmium preexposure at a level of dietary intake was shown to potentiate pulmonary inflammation on subsequent infection with respiratory syncytial virus (Hu et al. 2019). The body of evidence regarding cadmium's immunotoxicity is not entirely consistent; for example, a study of mice found that exposure to 300-ppm cadmium stimulated (rather than impaired) the memory response of lymphocytes to antigen (Koller and Roan 1980). Nevertheless, most studies have suggested that cadmium is generally immunotoxic (Cohen 2004).

We examined the prospective association of biomarkers of cadmium exposure with mortality related to influenza-related conditions using the U.S. National Health and Nutrition Examination Survey (NHANES) database. This would be the first such analysis of which we are aware.

Methods

Study Population

The present study used data from NHANES studies conducted between 1988 and 1994 (NHANES-III) and between 1999 and 2006 (continuous NHANES). We restricted continuous NHANES cycles through 2005–2006 because no mortality cases from influenza or pneumonia were reported in data from later cycles. The NHANES studies were not conducted between 1995 and 1998. NHANES, which is conducted by the National Center for Health Statistics (NCHS), provides nationally representative survey data on the

Address correspondence to Howard Hu, Department of Preventive Medicine, Keck School of Medicine University of Southern California, 2001 N. Soto St., Health Sciences Campus, Los Angeles, CA 90089-9013 USA. Telephone: (323) 865-0803. Email: howard.hu@med.usc.edu

Supplemental Material is available online (https://doi.org/10.1289/EHP7598).

The authors declare they have no actual or potential competing financial interests.

Received 3 June 2020; Revised 7 November 2020; Accepted 9 November 2020; Published 16 December 2020.

health and nutritional status of the noninstitutionalized U.S. population. Following a multistage complex sampling design, the survey includes data from interviews (demographic, socioeconomic, dietary, and health-related questions); examinations (medical and physiological measurements), and laboratory tests (biomarkers of exposures and outcomes). For the present study, participant inclusion criteria included *a*) being ≥45 years of age at the NHANES interview; *b*) having mortality status information available in the public-use linked mortality file as of the end of 2015 through linkage to the National Death Index (NDI); and *c*) having complete data on blood cadmium or urinary cadmium and core covariates [age, sex, race/ethnicity, education, body mass index (BMI), serum cholesterol, hypertension status, smoking status, and urinary creatinine (NHANES-III only)]. We used the 45-years-of-age cutoff because there were few mortality cases at <45 years of age. We excluded participants whose follow-up time was zero (6 in NHANES-III; 4 in NHANES 1999–2006). A total of 7,173 and 8,678 participants were used for the analysis in NHANES-III and NHANES 1999–2006, respectively. In NHANES-III, cadmium was measured in urine. In NHANES 1999–2006, cadmium was measured in blood and urine, but urinary cadmium was measured in one-third of the participants ($n = 2{,}846$). We examined only blood cadmium in NHANES 1999–2006 because the number of death cases from influenza or pneumonia in this subsample was too few ($n = 18$).

NHANES data collection procedures were approved by the NCHS Ethics Review Board, and participant written informed consent was obtained. All data used in this study are publicly available at https://github.com/um-mpeg/Cadmium-Influenza-Pneumonia-Mortality.

Mortality from Influenza or Pneumonia

The NCHS linked NHANES with death certificate records from the NDI (NCHS 2019), a centralized database compiled from state vital statistics offices. The linkage allows for access to data on mortality status, leading cause of death, and calculation of time from NHANES interview to death or end of follow-up. Causes of death are provided as categorized according to the *International Classification of Diseases, Ninth Revision, Clinical Modification* (ICD-9-CM; CDC 2013) codes (for deaths before 1999) and according to the *International Statistical Classification of Diseases and Related Health Problems, 10th Revision* (ICD-10; WHO 2016) codes (for deaths from 1999 to 2015). The public-use linked mortality file provides ICD-10–based underlying cause-of-death groups with a recode of all ICD-9–based mortality data. For the present study, we included mortality from influenza and pneumonia (ICD-10 codes J09–J18), which includes deaths from either influenza or pneumonia while excluding other chronic lower respiratory diseases (ICD-10 codes J40–J47). It should be noted that to reduce the risk of participant reidentification, the public-use linked mortality data were released after application of statistical disclosure limitation methods, including perturbation of follow-up time and underlying cause of death (NCHS 2020). Therefore, it is possible that the public-use linked mortality data could include synthetic influenza or pneumonia death cases and their follow-up times.

Blood and urinary cadmium were measured at the Environmental Health Sciences Laboratory of the Centers for Disease Prevention and Control (CDC) National Center for Environmental Health after confirmation of no background contamination in all collection and storage materials. Cadmium concentrations were measured by a simultaneous multielement atomic absorption spectrometer (Model SIMAA 6000l; Perkin-Elmer) with Zeeman background correction in NHANES-III and 1999–2002 and by an inductively coupled plasma–mass spectrometer (Model SCIEX 500; Perkin-Elmer) in subsequent continuous NHANES cycles. The limit of detection (LOD) for urinary cadmium was 0.01 μg/L and for blood cadmium, 0.2–0.3 μg/L. Cadmium concentrations below LODs were replaced with the LOD divided by the square root of 2.

Covariates

Sociodemographic factors [age, sex, race/ethnicity (non-Hispanic White, non-Hispanic Black, Mexican American, and other], education, and cigarette smoking (smoking status, pack-years) were collected with questionnaires at survey interviews. Education was selected as an indicator of socioeconomic status because it has less missing values than other indicators, such as the poverty:income ratio (PIR). We evaluated the residual confounding effect by PIR as a sensitivity analysis. Serum cotinine, a measure of environmental tobacco smoke exposure, was measured by isotope dilution–high-performance liquid chromatography/atmospheric pressure chemical ionization–tandem mass spectrometry. Markers of iron store (e.g., iron, total iron binding capacity, transferrin saturation, ferritin) that can influence cellular cadmium absorption were measured by an automated AAII-25 colorimetric method and an immunoradiometric method. BMI was calculated as weight in kilograms divided by height in meters squared. Urinary creatinine was measured using an enzymatic (creatinase) method.

Statistical Analysis

NHANES used a complex, stratified, multistage cluster sampling design to obtain a nationally representative sample of the noninstitutionalized U.S. civilian population. To account for the design, we used the survey package (version 4.0; https://www.rdocumentation.org/packages/survey/versions/4.0) in R (version 3.6.3; R Development Core Team). All analyses began with design-based univariate analyses using histograms, statistical summaries, and smoothing to examine outliers and general characteristics.

To account for urine dilution/hydration that can influence urinary biomarker concentrations, we used creatinine-corrected urinary cadmium [urinary cadmium divided by urinary creatinine concentrations (in micrograms per gram)], Recently, an improved method, covariate-adjusted standardization plus covariate adjustment for urinary creatinine, was recommended to reduce measurement error owing to urine dilution if there is an association between the outcome and urinary creatinine (O'Brien et al. 2016). We chose creatinine-corrected urinary cadmium as the primary exposure variable because urinary creatinine was not associated with the outcome of the present study, namely, mortality owing to influenza or pneumonia (hereafter called influenza/pneumonia) [with a hazard ratio (HR) of 1.0] and because it allows comparisons with previous NHANES studies of urinary cadmium and mortality (Menke et al. 2009). We conducted a sensitivity analysis using the improved approach (i.e., covariate-adjusted standardization plus covariate adjustment) and confirmed that the results were robust (Table S1).

To model the associations between biomarkers of cadmium and influenza/pneumonia mortality, we used survey-weighted Cox proportional hazards models (svycoxph in the R package survey), an extension of the common Cox model that incorporates complex survey design factors (clusters, strata, and sampling weights). We used attained age (i.e., age at influenza or pneumonia death, which was calculated as the sum of baseline age in months and follow-up time in months) as the time scale instead of follow-up time (time-on-study) because time-on-study as the time scale may misspecify the hazard function in longitudinal follow-up studies (Korn et al. 1997). We additionally adjusted for phase (NHANES-III, Phase 1 from October 1988 to October 1991, or Phase 2 from September

1991 to October 1994) or survey cycle (NHANES 1999–2000, 2001–2002, 2003–2004, or 2005–2006) to filter out any residual confounding by secular trends.

For older adults, age was top-coded at 90 (NHANES-III) or 85 y (NHANES 1999–2006) to protect the confidentiality of the survey participants (CDC/NCHS 1997, 2009). In other words, all adults ≥90 (or ≥85) years of age have an age variable value of 90 (or 85). Given that age-related mortality risks may not be uniform and generally increase with age even in this top-coded age category, confounding effects by age cannot be properly controlled for and may lead to bias other age-dependent covariates, including cadmium biomarkers. NHANES 1999–2006 provided the weighted mean age for participants aged 85 years and older: 88 y for NHANES 1999–2000 and NHANES 2003–2004; and 89 y for NHANES 2001–2002 and NHANES 2005–2006. We used the weighted mean age for each cycle instead of the top-coded age of 85 y in order to reduce potential age effects. NHANES-III data does not provide the weighted mean age for the top-coded age category. We chose 93 y based on the weighted mean ages reported for NHANES 1999–2006, and, so conservatively evaluated the association between cadmium and influenza/pneumonia mortality. As a sensitivity analysis, we conducted analyses after excluding participants in the top-coded age category.

HRs and 95% confidence intervals (CIs) were computed comparing the 80th percentile with the 20th percentile of the cadmium biomarker distributions fitted as a linear continuous term. The 80th and 20th percentiles and differences between the two were 1.20, 0.34, and 0.86 μg/g, respectively, for creatinine-corrected urinary cadmium in NHANES-III, and 0.80, 0.30, and 0.50 μg/L, respectively, for blood cadmium in NHANES 1999–2006. We also computed HRs and 95% CIs comparing the lowest vs. higher tertiles to better capture nonlinear dose–response relationships. Tests for linear trend across tertiles were conducted by fitting the tertiles as an ordinal variable (i.e., coded as 1, 2, and 3) in the models. We constructed four sequential models to evaluate for potential confounding: Model 1 was adjusted for sex, race/ethnicity, and phase (NHANES-III) or cycle (NHANES 1999–2006); Model 2 was further adjusted for education, BMI, serum total cholesterol, and hypertension; and Model 3 was further adjusted for smoking status and serum cotinine (log-transformed). In Model 4, we evaluated the associations only among never smokers. Models 3 and 4 facilitated the ability to disentangle cadmium from smoking, given that it is a major route of exposure to cadmium. We also repeated the analysis among ever smokers for the comparison by smoking status. Sensitivity analyses were conducted to assess for additional potential confounding related to variables for which the sample sizes were substantially smaller because of missing data. Thus, we repeated the analyses to evaluate for any potential residual confounding effects of PIR and pack-years of smoking. Another sensitivity analysis for additional adjustment for iron store markers (i.e., serum iron, total iron binding capacity, transferrin saturation, or ferritin) was conducted in the NHANES-III data. Adjustment for iron store was not conducted in the NHANES 1999–2006 data because iron store markers were measured only among children and women 12–59 years of age. In addition, we evaluated interaction between creatinine-corrected urinary cadmium and smoking status in NHANES-III on both multiplicative and additive scales. Interaction between blood cadmium and smoking status in NHANES 1999–2006 was not evaluated owing to the small number of mortality cases. To allow the additive-scale interaction, creatinine-corrected urinary cadmium was dichotomized at the median (0.653 μg/g) and smoking status was divided into never and ever smokers. The relative excess risk owing to interaction was computed as the measure of interaction on the additive scale (Knol and VanderWeele 2012).

We created adjusted cumulative hazard function plots for influenza/pneumonia mortality comparing high vs. low cadmium biomarker dichotomized at the median. To account for confounding in these plots, we used a counterfactual approach by comparing the counterfactual survivals that would have been observed if everyone had had high exposure vs. if everyone had had low exposure (Hernán and Robins 2020). Inverse probability weights (IPWs) for each individual were estimated by fitting logistic regression models with the binary cadmium biomarker as the outcome and the variables in Model 2 as the predictors and then stabilized. These stabilized IPWs were applied to obtaining adjusted cumulative functions independent of confounding.

A competing risk is an event in which the occurrence of a primary event of interest precludes the occurrence of the others and, therefore, can lead to invalid estimations for cause-specific events (Austin et al. 2016). A few statistical approaches have been suggested (Austin et al. 2016), but no approach is available that can accommodate both competing risks and the complex survey data. We used the Fine-Gray subdistribution hazard models (Fine and Gray 1999) for influenza/pneumonia vs. other deaths without the complex survey components to estimate what would have been HRs in the presence of competing risks. We used the crr() function (competing risks regression) in the R package cmprsk.

We tested for potential effect modification by age, sex, and race (non-Hispanic White, non-Hispanic Black). To evaluate effect modification, multiplicative interaction terms along with the main effects were included in regression models. Only linear cadmium biomarkers were used in this analysis owing to the small number of mortality cases.

Results

In NHANES-III, 7,173 participants were included, representing 66.6 million adults. The sample-weighted mean age and the proportion of females were 60.7 y (95% CI: 60.1, 61.4) and 53.1% (Table 1). With a median follow-up of 17.3 y, 141 participants died from influenza/pneumonia [unweighted incidence rate = 1.24 (95% CI: 1.04, 1.460 per 1,000 person-years; weighted incidence rate = 0.87 (95% CI: 0.87, 0.88) per 1,000 person-years]. The geometric mean, range, and percentage below the LOD value for urinary cadmium were 0.48 μg/L (95% CI: 0.45, 0.52), 0.01–16.6 μg/L, and 3.3%. In NHANES 1999–2006, 8,678 participants were included, representing 87.7 million adults. The sample-weighted mean age and the proportion of females were 59.6 y (95% CI: 59.1, 60.1) and 52.7%. During a median follow-up of 11.4 y, 56 participants died from influenza/pneumonia [unweighted incidence rate = 0.59 (95% CI: 0.44, 0.76) per 1,000 person-years; weighted incidence rate = 0.37 (95% CI: 0.36, 0.37) per 1,000 person-years]. The geometric mean, range, and percentage below the LOD value for blood cadmium were 0.46 μg/L (95% CI: 0.44, 0.47), 0.1–10.8 μg/L, and 12.5%.

Table 2 presents survey-weighted Cox regression results. In NHANES-III, after adjusting for sex, race/ethnicity, and phase (Model 1), creatinine-corrected urinary cadmium comparing the 80th vs. 20th percentiles was associated with an HR of 1.13 (95% CI: 1.02, 1.25; $p = 0.02$). The HR remained essentially unchanged and statistically significant with further adjustment for education, BMI, serum cholesterol, and hypertension [Model 2: HR = 1.15 (95% CI: 1.05, 1.26; $p = 0.002$)]. The HR decreased to 1.09 (95% CI: 0.94, 1.27; $p = 0.24$) after further adjustment for smoking status and serum cotinine (Model 3). However, when restricting the Model 2 analysis to never smokers, the HR comparing the 80th

Table 1. Survey-weighted characteristics of participants ≥45 years of age with mortality status and complete data on blood or urinary cadmium and core covariates.

Characteristics	NHANES-III	NHANES 1999–2006
Participants (*n*)	7,173	8,678
Influenza/pneumonia deaths (*n*)	141	56
Follow-up years [median (Q1–Q3)]	17.3 (8.8–23.2)	11.4 (9.4–13.9)
Mortality from influenza/pneumonia (per 1,000 person-years) [incident rate (95% CI)]		
Unweighted	1.24 (1.04, 1.46)	0.59 (0.44, 0.76)
Weighted	0.87 (0.87, 0.88)	0.37 (0.36, 0.37)
Continuous variables [mean (95% CI)]		
Urinary cadmium (μg/L)	0.48 (0.45, 0.52)[a]	—
Creatinine-corrected cadmium (μg/g)	0.58 (0.55, 0.60)[a]	—
Urinary creatinine (mg/L)	84.2 (81.7, 86.7)[a]	—
Blood cadmium (μg/L)	—	0.46 (0.44, 0.47)[a]
Age (y)	60.7 (60.1, 61.4)	59.6 (59.1, 60.1)
Body mass index (kg/m^2)	27.3 (27.1, 27.5)	28.7 (28.5, 28.9)
Serum total cholesterol (mg/dL)	222 (220, 223)	210 (209, 211)
Serum cotinine (mg/dL)	1.12 (0.92, 1.37)[b]	0.35 (0.30, 0.41)[b]
Categorical variables (%)		
Female	53.1	52.7
Race/ethnicity		
Non-Hispanic White	82.6	79.0
Non-Hispanic Black	8.2	8.8
Mexican American	3.0	4.3
Other	6.2	7.9
Education		
<High school	32.2	20.6
High school diploma	49.2	26.5
>High school	18.6	52.9
Poverty:income ratio		
<1.0	9.1	8.6
≥1.0	83.3	84.3
Missing	7.6	7.1
Smoking status		
Never	41.6	45.9
Former	36.7	34.6
Current	21.7	19.4
Hypertension	29.7	29.1

Note: Core covariates include age, sex, race/ethnicity, education, body mass index, serum total cholesterol, hypertension status, and phase (NHANES-III) or cycle (NHANES 1999–2006). Data are complete for all variables unless otherwise indicated. —, Not applicable; CI, confidence interval; NHANES, National Health and Nutrition Examination Survey; Q, quartile.
[a]Geometric means and 95% CIs are presented.
[b]Serum cotinine had 125 and 74 missing observations in NHANES-III and NHANES 1999–2006, respectively.

Table 2. Hazard ratios (95% CIs) of mortality from influenza/pneumonia comparing the 80th vs. the 20th percentile of the distribution of cadmium biomarker levels in the entire population and by smoking status.

	NHANES-III		NHANES 1999–2006	
	HR (95% CI)	*p* Value	HR (95% CI)	*p* Value
All				
Deaths/total (*n*/*N*)	141/7,173	—	56/8,678	—
Model				
1	1.13 (1.02, 1.25)	0.02	1.15 (0.98, 1.35)	0.09
2	1.15 (1.05, 1.26)	0.002	1.14 (0.96, 1.36)	0.15
3	1.09 (0.94, 1.27)	0.24	1.14 (0.94, 1.39)	0.18
Never smokers				
Deaths/total (*n*/*N*)	58/3,185	—	27/3,989	—
Model				
1	1.27 (1.10, 1.46)	0.001	1.59 (0.91, 2.78)	0.10
2	1.27 (1.12, 1.43)	0.0002	1.71 (0.95, 3.09)	0.07
3	1.26 (1.11, 1.44)	0.0004	1.71 (0.96, 3.06)	0.07
Ever smokers				
Deaths/total (*n*/*N*)	83/3,988	—	29/4,689	—
Model				
1	0.98 (0.75, 1.29)	0.91	1.13 (0.92, 1.38)	0.24
2	1.05 (0.87, 1.27)	0.61	1.14 (0.94, 1.39)	0.18
3	0.85 (0.63, 1.15)	0.28	1.03 (0.76, 1.39)	0.85
Participants with the top-coded age excluded[a]				
All				
Deaths/total (*n*/*N*)	137/7,104	—	42/8,308	—
Model				
1	1.13 (1.02, 1.25)	0.02	1.14 (0.96, 1.37)	0.13
2	1.15 (1.05, 1.26)	0.002	1.16 (0.95, 1.41)	0.14
3	1.09 (0.94, 1.27)	0.25	1.12 (0.87, 1.43)	0.38
Never smokers				
Deaths/total (*n*/*N*)	55/3,134	—	18/3,773	—
Model				
1	1.27 (1.10, 1.46)	0.001	Did not converge	—
2	1.27 (1.11, 1.44)	0.0003	Did not converge	—
3	1.26 (1.11, 1.44)	0.005	Did not converge	—
Ever smokers				
Deaths/total (*n*/*N*)	82/3,970	—	24/4,535	—
Model				
1	0.98 (0.75, 1.29)	0.89	1.14 (0.95, 1.37)	0.16
2	1.05 (0.86, 1.27)	0.63	1.17 (0.98, 1.39)	0.09
3	0.85 (0.62, 1.15)	0.29	1.10 (0.84, 1.44)	0.50

Note: Cadmium biomarker was fitted as a linear term. The 80th and 20th percentiles were 1.20 and 0.34 μg/g, respectively, for creatinine-corrected cadmium in NHANES-III; 0.80 and 0.30 μg/L, respectively, for blood cadmium in NHANES 1999–2006. Survey-weighted Cox proportional hazards models with attained age as the time scale were used to estimate HRs and 95% CIs. Model 1: adjusted for sex, race/ethnicity, and phase (NHANES-III) or cycle (NHANES 1999–2006). Model 2: further adjusted for education, body mass index, serum total cholesterol, hypertension status. Model 3: further adjusted for serum cotinine (log-transformed) and smoking status (only for all subjects). —, Not applicable; CI, confidence interval; HR, hazard ratio; NHANES, National Health and Nutrition Examination Survey.
[a]Age ≥90 y in NHANES-III ($n = 69$ excluded); age ≥85 y in NHANES 1999–2006 ($n = 370$ excluded).

with the 20th percentiles of cadmium was 1.27 (95% CI: 1.12, 1.43; $p = 0.002$). Further adjustment for serum cotinine in never smokers did not change the result. No significant association was found among ever smokers [Model 2: HR = 1.05 (95% CI: 0.87, 1.27; $p = 0.61$)]. HRs remained identical to corresponding HRs from the primary model in a sensitivity analysis with the exclusion of participants with the top-coded age of ≥90 y ($n = 69$ excluded). No significant interaction between creatinine-corrected urinary cadmium and smoking status was found on either multiplicative or additive scales (Table S2). The HRs comparing participants in the highest with the lowest tertiles were 2.29 (95% CI: 1.27, 4.11; $p_{trend} = 0.008$) in Model 2; and 1.59 (95% CI: 0.89, 2.84; $p_{trend} = 0.11$) in Model 3 (Table S3).

In NHANES 1999–2006, blood cadmium comparing the 80th with the 20th percentiles (equivalent to an increment of 0.50 μg/L) was associated with an HR of 1.15 (95% CI: 0.98, 1.35; $p = 0.09$) with adjustment for sex, race/ethnicity, and cycle (Model 1); 1.14 (95% CI: 0.96, 1.36; $p = 0.15$) with further adjustment for education, BMI, serum cholesterol, and hypertension (Model 2); and 1.14 (95% CI: 0.94, 1.39; $p = 0.18$) with further adjustment for smoking status and serum cotinine (Model 3) (Table 2). When the Model 2 analysis was restricted to never smokers, the HR for cadmium was 1.71 (95% CI: 0.95, 3.09; $p = 0.07$) and remained the same even with further adjustment for serum cotinine (Table 2). A relatively weak association was observed among ever smokers [Model 2: HR = 1.14 (95% CI: 0.94, 1.39; $p = 0.18$)]. In a sensitivity analysis after excluding participants ≥85 years of age ($n = 370$ excluded) whose true age was top-coded, HRs for the entire population were similar to the primary model (but with wider CIs), whereas positive HRs among ever smokers increased slightly. The models did not converge in never smokers owing to the small number of cases (18 cases). The HRs comparing participants in the second and third tertiles with those in the first tertile were 0.46 (95% CI: 0.23, 0.94) and 1.52 (95% CI: 0.79, 2.92) ($p_{trend} = 0.21$) in

Model 2 and 0.46 (95% CI: 0.22, 0.97) and 1.64 (95% CI: 0.78, 3.44) ($p_{trend} = 0.23$) in Model 3 (Table S3).

The adjusted cumulative hazard function plots suggest that the hazards of influenza/pneumonia mortality increased exponentially with age, starting from around age 70 y and a steeper increase in the high-exposure group compared with the low-exposure group was seen starting from around age 80 y (Figure 1). The cumulative hazard for the low blood cadmium group exceeded that for the high blood cadmium group at ~100 years of age in NHANES 1999–2006, but it seems to be because only a few participants remained at risk at that age.

In sensitivity analyses, the HRs remained unchanged with additional adjustments for either PIR or pack-years of smoking (Table S4). Further adjustment for iron store markers (i.e., serum iron, total iron binding capacity, transferrin saturation, or ferritin) in NHANES-III also did not change the HRs (Table S4).

We conducted subdistribution hazard models for both influenza/pneumonia and noninfluenza/pneumonia deaths to evaluate potential competing risks. We also compared cause-specific HRs between influenza/pneumonia and noninfluenza/pneumonia deaths. Creatinine-corrected urinary cadmium in NHANES-III had similar associations on the relative incidence and cause-specific hazard of influenza/pneumonia death as on the relative incidence and cause-specific hazard of noninfluenza/pneumonia death, indicating the impact of competing risks to be minimal (Table S5). Blood cadmium had a slightly less pronounced association on the incidence of influenza/pneumonia death compared with the association on the incidence of noninfluenza/pneumonia death, although cause-specific hazards for both outcomes were similar (Table S5).

An evaluation of effect modification showed that there was no clear evidence of effect modification by age, sex, or race (non-Hispanic White vs. non-Hispanic Black) for both creatinine-corrected urinary cadmium in NHANES-III and blood cadmium in NHANES 1999–2006, although a substantially elevated HR was observed among very old adults ≥90 years of age in NHANES-III [Model 2: HR = 1.63 (95% CI: 0.89, 2.98; $p = 0.12$)] (Figure 2; Table S6).

Discussion

This is the first report of which we are aware showing that cadmium body burden measured as urinary concentrations may be associated with higher risk of mortality from influenza or pneumonia among older adults representative of the general U.S. population. This association remains statistically significant even among never smokers. Although power was limited by the small number of mortality cases, we also found a trend toward increased mortality with higher levels of blood cadmium. The stronger association of mortality with urinary vs. blood cadmium levels suggests that the primary risk factor is cadmium burden that has accumulated over time vs. recent cadmium exposure. Given the very low excretion rate of cadmium, urinary cadmium levels have been shown to best reflect chronic, long-term exposure, whereas blood cadmium levels are more responsive to acute, recent exposures (Vacchi-Suzzi et al. 2016).

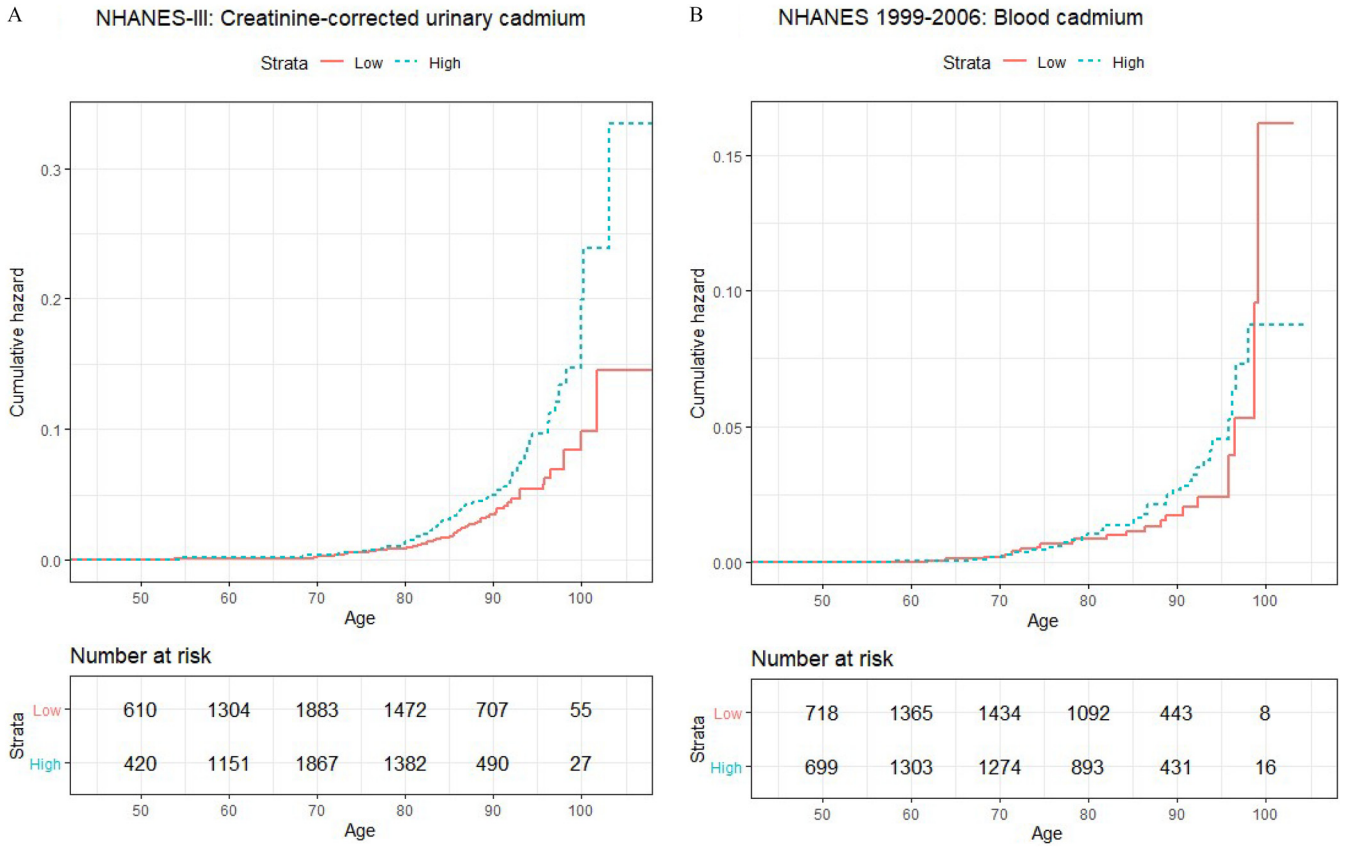

Figure 1. Adjusted cumulative hazard plots for mortality from influenza and pneumonia by cadmium biomarker in (A) NHANES-III and (B) NHANES 1999–2006. Cadmium biomarker was dichotomized into low and high at the median (0.653 μg/g for creatinine-corrected cadmium in NHANES-III; 0.48 μg/L for blood cadmium in NHANES 1999–2006). To account for confounding, adjusted cumulative hazard function estimates were computed using inverse probability weights that were estimated by fitting logistic regression models with the binary cadmium biomarker as the outcome and age, sex, race/ethnicity, education, body mass index, serum total cholesterol, hypertension status, and phase (NHANES-III) or cycle (NHANES 1999–2006) as the predictors and then stabilized. Note: NHANES, National Health and Nutrition Examination Survey.

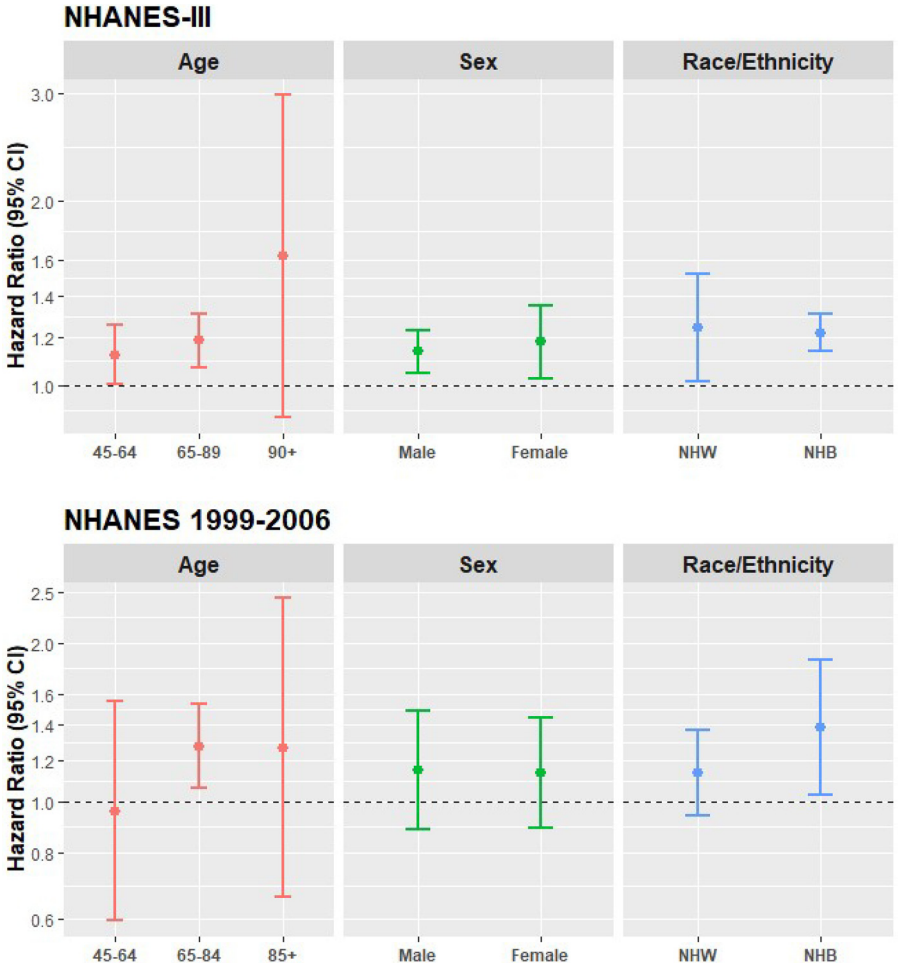

Figure 2. Hazard ratios (95% CIs) of mortality from influenza/pneumonia comparing the 80th vs. the 20th percentile of the distribution of cadmium biomarker levels by participant subgroup. Cadmium biomarker was fitted as a linear term. The 80th and 20th percentiles were 1.20 and 0.34 μg/g, respectively, for creatinine-corrected cadmium in NHANES-III; 0.80 and 0.30 μg/L, respectively, for blood cadmium in NHANES 1999–2006. Survey-weighted Cox proportional hazards models with attained age as the time scale were used to estimate hazard ratios and 95% CIs. All models were adjusted for sex, race/ethnicity, education, body mass index, serum cholesterol, hypertension, and phase (NHANES-III) or cycle (NHANES 1999–2006). Note: CI, confidence interval; NHANES, National Health and Nutrition Examination Survey; NHB, non-Hispanic Black; NHW, non-Hispanic White. See Table S6 for corresponding numeric data and $p_{\text{interaction}}$-values.

There are a variety of reasons why cadmium burden may predispose individuals to worse pulmonary disease from respiratory infections, including evidence that cadmium potentiates cytokine production (Låg et al. 2010) and lung inflammation, impairs macrophage-mediated immune function (Koller 1998), disrupts tight junction integrity in human airway epithelial tissue at the level of the air–liquid–interface airway (Cao et al. 2015), and potentiates pulmonary inflammation on subsequent infection with respiratory syncytial virus (Hu et al. 2019). Cadmium is known to accumulate in human lung tissues (Hassan et al. 2014). Cadmium binds to the sulfhydryl group of thiols, such as glutathione and metallothionine, and interferes with the antioxidant defense system, leading to an altered redox balance and oxidative stress (Moulis 2010). Local accumulation of cadmium in the lungs has been linked to impaired innate immunity, which can enhance vulnerability to viral infections (Chandler et al. 2019; Hu et al. 2019). Blood levels of cadmium have been found to be associated with a reduced forced expiratory volume in 1 s:forced vital capacity (FEV_1/FVC) ratio (<0.7) in a cross-section analysis of NHANES 2007–2010 data (Rokadia and Agarwal 2013), and urinary cadmium levels have been shown to be associated with reduced lung function (FEV_1 and FEV_1/FVC ratio) in a cohort study of aging men, regardless of smoking status (Lampe et al. 2008). These effects, in turn, may predispose individuals to worse pulmonary infectious disease outcomes.

We evaluated whether subpopulations are more susceptible to cadmium burden-related mortality risk from influenza and pneumonia but found no clear differences in the associations by age, sex, or race (Figure 2; Table S6). However, these findings should be interpreted with caution because the number of cases is small.

Overall, these findings suggest that cadmium may play a role in predisposing people to influenza-related pneumonia mortality no matter whether the sources are from cigarette smoking or contaminated food intake, the major route of exposure for nonsmokers (Satarug 2018). Given the intense worldwide spread of COVID-19, it will be important to determine whether cadmium burden is a risk factor for worse outcomes in this disease. As reflected by the most recent NHANES data (2007–2012) on cadmium levels, average values for estimated dietary cadmium consumption in the United States remains substantial, with children ≤10 years of age among those with the highest cadmium intakes on a body weight basis (Kim et al. 2018). Environmental health policies may be needed to monitor and reduce population exposures to cadmium, which include exposure via cadmium emissions into air, soil, water, and sewage from mining, smelting, and various other industries (e.g. via NiCd batteries, plating, pigments, plastics) and their

eventual uptake in common foods (ATSDR 2012). Food groups estimated to contribute most to total cadmium intake among NHANES participants (Kim et al. 2018) are cereals and bread (34%), leafy vegetables (20%), potatoes (11%), legumes and nuts (7%), and stem/root vegetables (6%), with an average total dietary cadmium consumption of 4.63 μg/d.

Finally, if cadmium burden does adversely impact lung infections, there are some secondary prevention strategies that could potentially mitigate the impact. A number of studies have demonstrated the ability of *N*-acetyl cysteine, a precursor of reduced glutathione and a well-known anti-oxidant, to reduce cadmium toxicity in cultured liver and renal cells (Luo et al. 2013; Odewumi et al. 2011; Wang et al. 2014). Further, in an experimental study of human lung cells exposed to cadmium chloride, treatment with *N*-acetyl cysteine was found to improve viability from 44.5% to 84.1%, with concomitant reductions in the numbers of cytokines that were up- and down-regulated (Odewumi et al. 2016).

Our study has several limitations. First, the present study used public-use linked mortality data instead of restricted-use linked mortality data. Data perturbation was done in the public-use linked mortality data by adding random noise to two confidential elements—follow-up time and underlying cause of death—to reduce reidentification risk to participants, which may cause bias in Cox proportional hazards models (NCHS 2020). Random noise added to follow-up time and/or underlying cause of death is expected to result in nondifferential misclassification of the outcome (i.e., follow-up time or status of influenza or pneumonia) and seems likely to have biased results toward the null. Therefore, this potential bias should be acknowledged when public-use linked mortality data is used.

Second, accurate attained age during follow-up could not be calculated for older participants because their ages were top-coded as ≥90 y for those ≥90 years of age in NHANES-III and ≥85 y for those ≥85 years of age in NHANES 1999–2006 in the public-use NHANES cross-sectional data and because the public-use mortality file contains follow-up time since interview/exam as the sole survival time-related variables. Because no additional information necessary for calculating accurate attained age is available in these two public NHANES data sources, this could lead to insufficient adjustment for confounding by age, but the direction of any bias is unknown. To address this issue, we conducted sensitivity analyses with the exclusion of those participants with a top-coded age. The sensitivity analyses produced results largely consistent with the main analyses, which could have suffered from some degree of residual confounding, suggesting that potential bias from the inaccurately calculated attained age was likely minimal. The sensitivity analysis for NHANES-III data produced results that were more consistent, as expected, given that the higher cutoff for top-coding and the longer follow-up meant that the analysis included relatively more participants for whom age was recorded accurately and who are then known to have attained a high age.

Third, it is known that the ICD-10 codes for influenza-related mortality (ICD-10 codes J10 and J11) cannot capture all cases because tests for influenza are not done for most adult patients and because many influenza-infected patients die from secondary bacterial infections or underlying chronic diseases (Thompson et al. 2009). If individuals with higher cadmium body burden were more likely to die from such secondary causes of death, our reported HRs would have been underestimated.

Fourth, the NHANES 1999–2006 analysis may be underpowered owing to the small number of cases. This also precluded us from including urinary cadmium, which was measured in only one-third of the participants. Nonetheless, the present study is the first population-based prospective analysis of cadmium burden and mortality from influenza/pneumonia, with long follow-up (up to 27 y) and high quality data obtained using rigorous protocols. Finally, although we conducted competing risk analyses that supported that the reported HRs may be independent of competing risks, these estimates should be interpreted with caution because the complex survey components were not incorporated in the analysis.

In conclusion, the present study suggests that a higher cadmium burden is associated with higher mortality from influenza/pneumonia. These findings were robust even among never smokers, suggesting that cadmium burden is a risk factor independent of smoking. Given the experimental evidence indicating that cadmium's mode of pulmonary toxicity is likely through immunotoxicity and amplifying inflammation (Cohen 2004), it is possible that higher cadmium burdens may worsen outcomes from COVID-19 infections, for which evidence is building that the main mechanism involves marked amplification of inflammation (Fu et al. 2020). Conversely, a potential intervention such as *N*-acetyl cysteine, which has been shown in animal studies to reduce the lung's inflammatory response to cadmium, may blunt the impact of COVID-19. Research is needed that directly focuses on cadmium burden as a risk factor for worse pulmonary COVID-19 outcomes, and, potentially, the ability for *N*-acetyl cysteine to mitigate such outcomes.

Acknowledgments

The authors thank J. Siren for his assistance in advancing our team's collaboration. This work was supported by a Rapid Response Pilot Project from the University of Michigan (P30 ES017885) to S.K.P., H.H. and C.S. was supported by a grant from the National Institutes of Health/National Institute of Environmental Health Sciences (K23-ES030725).

Analytical data and R codes are available at https://github.com/um-mpeg/Cadmium-Influenza-Pneumonia-Mortality.

References

ATSDR (Agency for Toxic Substances and Disease Registry). 2012. Toxicological Profile for Cadmium. https://www.atsdr.cdc.gov/toxprofiles/TP.asp?id=48&tid=15 [accessed 17 November 2020].

Austin PC, Lee DS, Fine JP. 2016. Introduction to the analysis of survival data in the presence of competing risks. Circulation 133(6):601–609, PMID: 26858290, https://doi.org/10.1161/CIRCULATIONAHA.115.017719.

Cao X, Lin H, Muskhelishvili L, Latendresse J, Richter P, Heflich RH. 2015. Tight junction disruption by cadmium in an in vitro human airway tissue model. Respir Res 16(1):30, PMID: 25851441, https://doi.org/10.1186/s12931-015-0191-9.

CDC/NCHS (Centers for Disease Control and Prevention/National Center for Health Statistics). 1997. *The Third National Health and Nutrition Examination Survey (NHANES III), 1A. Interview & Exam Components (July 1997).* https://www.cdc.gov/nchs/nhanes/nh3data.htm [accessed 8 August 2020].

CDC/NCHS. 2009. National Health and Nutrition Examination Survey 1999–2000 Data Documentation, Codebook, and Frequencies: Demographic Variables & Sample Weights (DEMO). https://wwwn.cdc.gov/Nchs/Nhanes/1999-2000/DEMO.htm [accessed 8 August 2020].

CDC (Centers for Disease Control and Prevention). 2013. *International Classification of Diseases, Ninth Revision, Clinical Modification (ICD-9-CM).* http://www.cdc.gov/nchs/icd/icd9cm.htm [accessed 17 November 2020].

Chandler JD, Hu X, Ko EJ, Park S, Fernandes J, Lee YT, et al. 2019. Low-dose cadmium potentiates lung inflammatory response to 2009 pandemic H1N1 influenza virus in mice. Environ Int 127:720–729, PMID: 30999129, https://doi.org/10.1016/j.envint.2019.03.054.

Cohen MD. 2004. Pulmonary immunotoxicology of select metals: aluminum, arsenic, cadmium, chromium, copper, manganese, nickel, vanadium, and zinc. J Immunotoxicol 1(1):39–69, PMID: 18958639, https://doi.org/10.1080/15476910490438360.

Fine JP, Gray RJ. 1999. A proportional hazards model for the subdistribution of a competing risk. J Am Stat Assoc 94(446):496–509, https://doi.org/10.1080/01621459.1999.10474144.

Fu Y, Cheng Y, Wu Y. 2020. Understanding SARS-CoV-2-mediated inflammatory responses: from mechanisms to potential therapeutic tools. Virol Sin 35(3):266–271, PMID: 32125642, https://doi.org/10.1007/s12250-020-00207-4.

Hassan F, Xu X, Nuovo G, Killilea DW, Tyrrell J, Da Tan C, et al. 2014. Accumulation of metals in GOLD4 COPD lungs is associated with decreased CFTR levels. Respir Res 15(1):69, PMID: 24957904, https://doi.org/10.1186/1465-9921-15-69.

Hernán MA, Robins JM. 2020. 17. Causal survival analysis. In: *Causal Inference: What If. R and Stata Code for Exercises*. Boca Raton: Chapman & Hall/CRC. https://remlapmot.github.io/cibookex-r/ [accessed 17 November 2020].

Hu X, Kim KH, Lee Y, Fernandes J, Smith MR, Jung YJ, et al. 2019. Environmental cadmium enhances lung injury by respiratory syncytial virus infection. Am J Pathol 189(8):1513–1525, PMID: 31108100, https://doi.org/10.1016/j.ajpath.2019.04.013.

Kim K, Melough MM, Vance TM, Noh H, Koo SI, Chun OK. 2018. Dietary cadmium intake and sources in the US. Nutrients 11(1):2, PMID: 30577418, https://doi.org/10.3390/nu11010002.

Knol MJ, VanderWeele TJ. 2012. Recommendations for presenting analyses of effect modification and interaction. Int J Epidemiol 41(2):514–520, PMID: 22253321, https://doi.org/10.1093/ije/dyr218.

Koller LD. 1998. Cadmium. In: *Immunotoxicology of Environmental and Occupational Metals*. Zelikoff JT, Thomas PT, eds. London, UK: Taylor and Francis, 41–61.

Koller LD, Roan JG. 1980. Effects of lead, cadmium and methylmercury on immunological memory. J Environ Pathol Toxicol 4(5–6):47–52, PMID: 7217859.

Korn EL, Graubard BI, Midthune D. 1997. Time-to-event analysis of longitudinal follow-up of a survey: choice of the time-scale. Am J Epidemiol 145(1):72–80, PMID: 8982025, https://doi.org/10.1093/oxfordjournals.aje.a009034.

Låg M, Rodionov D, Øvrevik J, Bakke O, Schwarze PE, Refsnes M. 2010. Cadmium-induced inflammatory responses in cells relevant for lung toxicity: expression and release of cytokines in fibroblasts, epithelial cells and macrophages. Toxicol Lett 193(3):252–260, PMID: 20105457, https://doi.org/10.1016/j.toxlet.2010.01.015.

Lampe BJ, Park SK, Robins T, Mukherjee B, Litonjua AA, Amarasiriwardena C, et al. 2008. Association between 24-hour urinary cadmium and pulmonary function among community-exposed men: the VA Normative Aging Study. Environ Health Perspect 116(9):1226–1230, PMID: 18795167, https://doi.org/10.1289/ehp.11265.

Luo YH, Wu SB, Wei YH, Chen YC, Tsai MH, Ho CC, et al. 2013. Cadmium-based quantum dot induced autophagy formation for cell survival via oxidative stress. Chem Res Toxicol 26(5):662–673, PMID: 23617821, https://doi.org/10.1021/tx300455k.

Menke A, Muntner P, Silbergeld EK, Platz EA, Guallar E. 2009. Cadmium levels in urine and mortality among U.S. adults. Environ Health Perspect 117(2):190–196, PMID: 19270787, https://doi.org/10.1289/ehp.11236.

Moulis JM. 2010. Cellular mechanisms of cadmium toxicity related to the homeostasis of essential metals. Biometals 23(5):877–896, PMID: 20524046, https://doi.org/10.1007/s10534-010-9336-y.

NCHS (National Center for Health Statistics). 2019. *The Linkage of National Center for Health Statistics Survey Data to the National Death Index – 2015 Linked Mortality File (LMF): Methodology Overview and Analytic Considerations*. Data Release Date: 6 November 2017. https://www.cdc.gov/nchs/data/datalinkage/LMF2015_Methodology_Analytic_Considerations.pdf [accessed 17 November 2020].

NCHS. 2020. 2015 Public-use linked files. https://www.cdc.gov/nchs/data-linkage/mortality-public.htm [accessed 12 October 2020].

O'Brien KM, Upson K, Cook NR, Weinberg CR. 2016. Environmental chemicals in urine and blood: improving methods for creatinine and lipid adjustment. Environ Health Perspect 124(2):220–227, PMID: 26219104, https://doi.org/10.1289/ehp.1509693.

Odewumi CO, Badisa VLD, Le UT, Latinwo LM, Ikediobi CO, Badisa RB, et al. 2011. Protective effects of N-acetylcysteine against cadmium-induced damage in cultured rat normal liver cells. Int J Mol Med 27(2):243–248, PMID: 21125209, https://doi.org/10.3892/ijmm.2010.564.

Odewumi CO, Latinwo LM, Ruden ML, Badisa VLD, Fils-Aime S, Badisa RB. 2016. Modulation of cytokines and chemokines expression by NAC in cadmium chloride treated human lung cells. Environ Toxicol 31(11):1612–1619, PMID: 26138014, https://doi.org/10.1002/tox.22165.

Rokadia HK, Agarwal S. 2013. Serum heavy metals and obstructive lung disease: results from the National Health and Nutrition Examination Survey. Chest 143(2):388–397, PMID: 22911427, https://doi.org/10.1378/chest.12-0595.

Satarug S. 2018. Dietary cadmium intake and its effects on kidneys. Toxics 6(1):15, PMID: 29534455, https://doi.org/10.3390/toxics6010015.

Thompson WW, Moore MR, Weintraub E, Cheng PY, Jin X, Bridges CB, et al. 2009. Estimating influenza-associated deaths in the United States. Am J Public Health 99(suppl 2):S225–S230, PMID: 19797736, https://doi.org/10.2105/AJPH.2008.151944.

Vacchi-Suzzi C, Kruse D, Harrington J, Levine K, Meliker JR. 2016. Is urinary cadmium a biomarker of long-term exposure in humans? A review. Curr Environ Health Rep 3(4):450–458, PMID: 27696280, https://doi.org/10.1007/s40572-016-0107-y.

Wang J, Zhu H, Liu X, Liu Z. 2014. N-acetylcysteine protects against cadmium-induced oxidative stress in rat hepatocytes. J Vet Sci 15(4):485–493, PMID: 25234327, https://doi.org/10.4142/jvs.2014.15.4.485.

WHO (World Health Organization). 2016. *International Statistical Classification of Diseases and Related Health Problems, 10th Revision*. http://apps.who.int/classifications/icd10/browse/2016/en [accessed 17 November 2020].